

A new species of *Micropterix* Hübner, 1825 from the Orobian Alps (Italy) (Lepidoptera, Micropterigidae)

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Abstract. *Micropterix gaudiella* Zeller & Huemer, **sp. n.** is described from the southern part of the Orobian Alps (Piedmont, Italy) and compared with its likely closest relatives *Micropterix isobasella* Staudinger, 1871 and *Micropterix stuebneri* Zeller, Werno & Kurz, 2013. The new species is well characterized by its wing pattern and colour and by structures of the male genitalia. The species status is furthermore supported by molecular data of the DNA barcode region. The distance to its nearest neighbour *Micropterix schaefferi* Heath, 1975 is 2.65%. *M. gaudiella* is the seventh species of the genus *Micropterix* Hübner, 1825 probably endemic to the Alps.

Introduction

The European fauna of *Micropterigidae* has recently gained increasing attention, reflected by faunistic reviews and several taxonomic papers (Corley 2007; Zeller-Lukashort et al. 2007; Thierry and Nel 2012; Zeller-Lukashort et al. 2013). The actual species inventory now seems well advanced as new species are rarely found and usually originate from insufficiently explored Mediterranean countries, with only two remarkable exceptions from the Central European Alps within the last four decades (Heath and Kaltenbach 1984; Kurz et al. 2004). Sampling of an unidentified *Micropterix* in the Italian Alps (Pizzo Arero, Orobian Alps, Piedmont, Italy) by PH and colleagues in June and July 2013 and 2014 thus came as a surprise. Although only few females were available in first hand, phenotypic appearance and the DNA barcode did not match any hitherto described species from the region. It was therefore decided to search for additional samples and we finally succeeded in collecting a large series of the species in summer 2014 including the male sex. Subsequent analysis of male genitalic characters supported the recognition of a new species, which is described here.

Material and methods

Our study is based on almost 277 specimens of the new *Micropterix* species and uncounted material of all European congeners. The type material is only partially set whereas several samples were only spread and dried immediately after collecting to ensure sufficient quality of DNA samples. Genitalia preparations followed standard techniques used for the family Micropterigidae (Zeller-Lukashort et al. 2007). Photographs of the adults were taken with an Olympus SZX 10 binocular microscope and an Olympus E-3 digital camera and developed using the software HELICON FOCUS 4.3, ADOBE PHOTOSHOP CS4 and LIGHTROOM 2.3. DNA barcode sequences are based on a 658 base-pair long segment of the mitochondrial COI gene (cytochrome c oxidase 1). DNA samples (dried legs) were prepared according to the prescribed standards (deWaard et al. 2008). Present authors and associated colleagues tried to obtain DNA barcodes of the majority of European Micropterigidae. Legs from 379 specimens belonging to 57 species of *Micropterix* have so far been processed at the Canadian Centre for DNA Barcoding (CCDB, Biodiversity Institute of Ontario, University of Guelph) to obtain DNA barcodes (BOLD 2015) using the standard high-throughput protocol described in Ivanova et al. (2006) and deWaard et al. (2008). DNA sequencing resulted in a barcode fragment of 658 bp for a total of 149 specimens and 29 species (BOLD 2015), partially published earlier by Lees et al. (2010). Ninety-one sequences belonging to 25 species are treated in this study and enable delimitation of the new species. Details of successfully sequenced voucher specimens including complete voucher data and images can be accessed in the Barcode of Life Data Systems: public dataset “Lepidoptera of the Alps - Micropterix [DS-LEALMIC]” (BOLD 2015; Ratnasingham and Hebert 2007). Degrees of intra- and interspecific variation in the DNA barcode fragment were calculated under Kimura 2-parameter (K2P) model of nucleotide substitution using analytical tools in BOLD Systems v3.0 (BOLD 2015). A neighbour-joining tree of DNA barcode data of European taxa was constructed using MEGA 5 (Tamura et al. 2011) under the K2P model for nucleotide substitutions.

The morphology of the new species is compared with similar species from the Alps and also from other regions of Europe (Kurz and Kurz 2015). We consequently build on the important identification treatments by Heath (1987), Kozlov (1989, 1990a, b) and Zeller-Lukashort et al. (2007). For more information about collection sites, preparation techniques and data archive of *Micropterix* spp. see Zeller-Lukashort et al. (2007).

Abbreviations of private and institutional collections:

MBCG	Italy, Bergamo, Museo di Scienze Naturali “Enrico Caffi”;
RCTM	Research Collection Toni Mayr, Feldkirch, Austria;
RCCZ	Research Collection Christof Zeller, Thalgau, Austria;
RCNP	Research Collection Norbert Pöll, Bad Ischl, Austria;
RCSO	Research Collection Siegfried Ortner, Bad Ischl, Austria;
TLMF	Tiroler Landesmuseum Ferdinandeum, Innsbruck, Austria.

Results

Checklist of European *Micropterix*

The species listed below occur within Europe (Karsholt 2015, Kurz and Kurz 2015), presented in an order considered to reflect morphological relationships among the species as suggested by Kurz

et al. (2015). Many of the morphological characters of *Micropterix* have been found to be more or less similar in smaller or greater groups of species and it has been assumed that similarities or synapomorphies of the male genitalia indicate a closer relationship. Based on a specific character matrix several characters have been selected and attributed as plesiomorphic or apomorphic (but see Kurz et al. 2015).

- Micropterix mansuetella* Zeller, 1844
- Micropterix amsella* Heath, 1975
- Micropterix calthella* (Linnaeus, 1761)
- Micropterix isobasella* Staudinger, 1871
- Micropterix stuebneri* Zeller, Werno & Kurz, 2013
- Micropterix gaudiella* sp. n.
- Micropterix granatensis* Heath, 1981
- Micropterix aglaella* (Duponchel, 1838)
- Micropterix wockei* Staudinger, 1870
- Micropterix aureatella* (Scopoli, 1763)
- Micropterix herminiella* Corley, 2007
- Micropterix aruncella* (Scopoli, 1763)
- Micropterix corcyrella* Walsingham, 1919
- Micropterix lakoniensis* Heath, 1985
- Micropterix kardamylensis* Rebel, 1903
- Micropterix igaloensis* Amsel, 1951
- Micropterix cassinella* Kurz, Kurz & Zeller, 2010
- Micropterix klimeschi* Heath, 1973
- Micropterix completella* Staudinger, 1871
- Micropterix tunbergella* (Fabricius, 1787)
- Micropterix sicanella* Zeller, 1847
- Micropterix cypriensis* Heath, 1985
- Micropterix aureoviridella* (Höfner, 1898)
- Micropterix maschukella* Alphéraky, 1876
- Micropterix facetella* Zeller, 1850
- Micropterix jeanneli* Viette, 1949
- Micropterix renatae* Kurz, Kurz & Zeller-Lukashort, 1997
- Micropterix minimella* Heath, 1973
- Micropterix italica* Heath, 1981
- Micropterix ertella* Walsingham, 1919
- Micropterix uxoria* Walsingham, 1919
- Micropterix paykullella* (Fabricius, 1794)
- Micropterix garganoensis* Heath, 1960
- Micropterix imperfectella* Staudinger, 1859
- Micropterix tuscaniensis* Heath, 1960
- Micropterix hartigi* Heath, 1981
- Micropterix allionella* (Fabricius, 1794)
- Micropterix trifasciella* Heath, 1965

- Micropterix rothenbachii* Frey, 1856
Micropterix huemeri Kurz, Kurz & Zeller-Lukashort, 2004
Micropterix ibericella Caradja, 1920
Micropterix zangheriella Heath, 1963
Micropterix schaefferi Heath, 1975
Micropterix emiliensis Viette, 1950
Micropterix osthelderi Heath, 1975
Micropterix trinakriella Kurz, Zeller-Lukashort & Kurz, 1997
Micropterix vulturensis Heath, 1981
Micropterix rablensis Zeller, 1868
Micropterix myrtetella Zeller, 1850
Micropterix croatica Heath & Kaltenbach, 1984
Micropterix fenestrellensis Heath & Kaltenbach, 1984

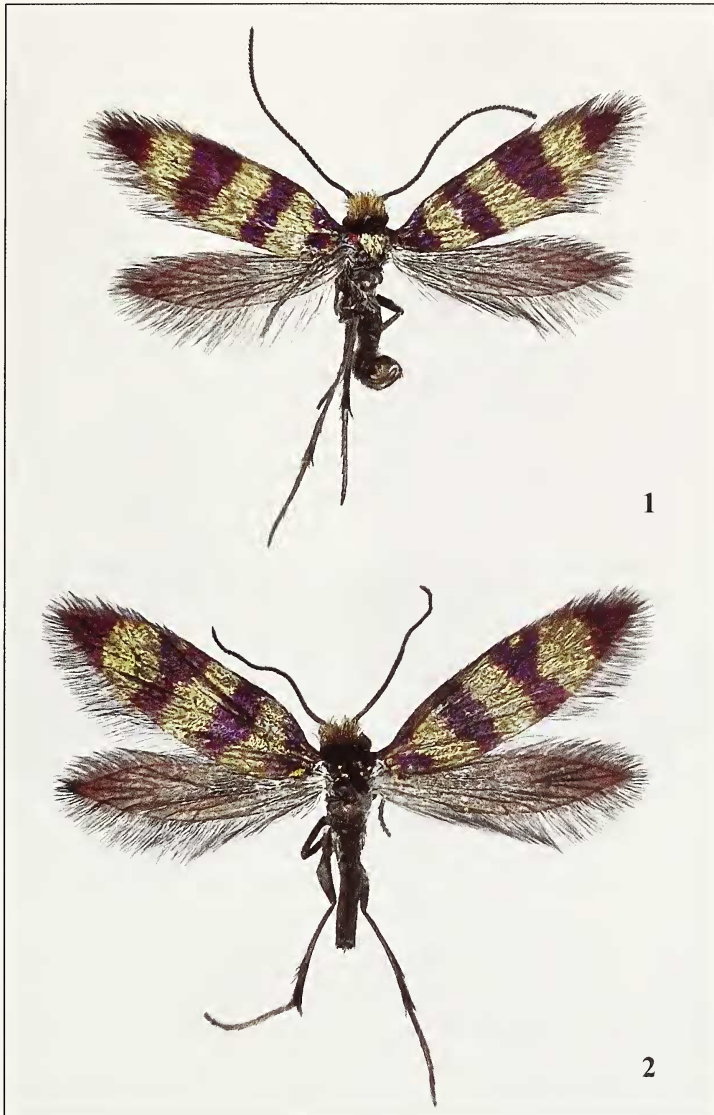
Taxonomic part

Micropterix gaudiella Zeller & Huemer, sp. n.

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Material. Holotype ♂: Italia sept., Bergamo, Alpi Orobie Rifugio Ca d'Arera 1600 m 9°47,8'E, 45°55,07'N 25.vi.2014, leg. Huemer TLMF 2014-006 (TLMF), label with identification numbers CZ-Z30577, label "DNA BARCODE TLMF Lep14851" and red label "HOLOTYPE of *Micropterix gaudiella* Zeller & Huemer". – Paratypes: 4 ♀, same locality as holotype, but 16.vii.2013, leg. Massaro M. (MBCG), labels with identification numbers CZ-Z30771, CZ-Z30774–CZ-Z30776; 3 ♂, 2 ♀, same locality as holotype, but 17.vii.2014, leg. Siegfried Ortner (RCSO), labels with identification numbers CZ-Z29417, CZ-Z29459–CZ-Z29462; 40 ♂, 22 ♀, same data as holotype, but leg. Norbert Pöll (RCNP), labels with identification numbers CZ-Z29463–CZ-Z29524; 69 ♂, 26 ♀, same data as holotype, but leg. Mayr Toni (RCTM), labels with identification numbers CZ-Z29360–CZ-Z29369, CZ-Z29371–CZ-Z29416, CZ-Z29418–CZ-Z29445, CZ-Z29446 and AP: MK-1072, CZ-Z29448–CZ-Z29457; 76 ♂, 15 ♀, same data as holotype (TLMF), labels with identification numbers CZ-Z29330–CZ-Z29347, CZ-Z30561–CZ-Z30576, CZ-Z30578–CZ-Z30580, CZ-Z30582–CZ-Z30623, CZ-Z30625–CZ-Z30631, CZ-Z30633–CZ-Z30634, CZ-Z30636–CZ-Z30638; 16 ♂, same locality as holotype, but 23.vi.2014, labels with identification numbers CZ-Z29314–CZ-Z29327, CZ-Z29328 and AP-Nr 1/2014, CZ-Z29329; 1 ♂, same data as holotype (TLMF), label with identification number CZ-Z30581, label "DNA BARCODING TMLF Lep14853"; 1 ♀, same data as holotype (TLMF), label with identification number CZ-Z30624, label "DNA BARCODING TMLF Lep14852"; 1 ♀, same locality as holotype, but 25.vi.2013, leg. Huemer (TLMF), label with identification number CZ-Z29359, label "DNA BARCODING TMLF Lep09987". All 276 paratypes bear red label "PARATYPE of *Micropterix gaudiella* Zeller & Huemer".

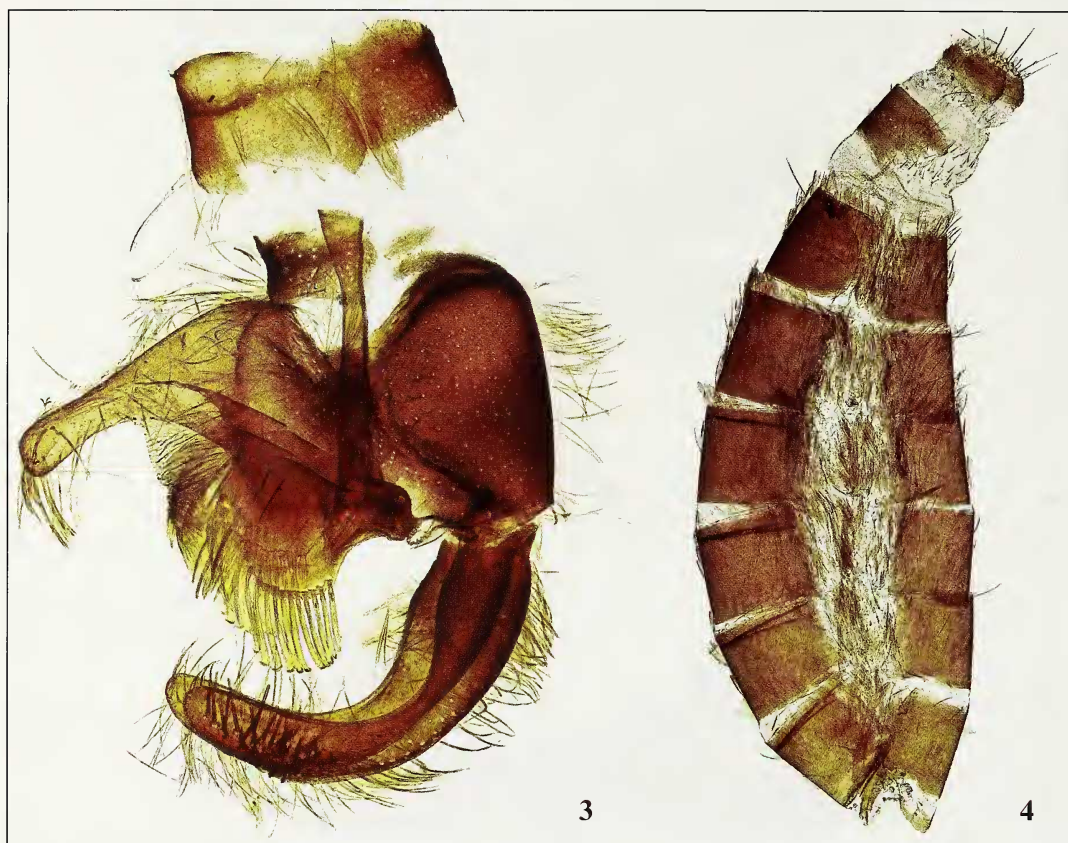
Description. Adult (Figs 1, 2). Forewing length 3.5–3.9 mm (♂, n=206), 4.0–4.4 mm (♀, n=71). Head black-brown; vestiture of hair-like scales on the head, yellow to dirty yellow; antennae dark brown, respectively 3/4 (♂) and 1/2 (♀) of forewing length; thorax and tegulae violet with golden reflection; forewing dull bluish with conspicuously broad and mostly bronzy golden markings: a trapeziform fascia at 1/4, at 1/2 a moderately broad straight fascia across the whole wing width, inwardly oblique, at 3/4 a broad, variably shaped, inwardly convex fascia; fringe bronzy golden, basally bluish violet; hindwing bronzy golden, apically tinged purplish; fringe bronzy golden; legs and abdomen brown, golden shining.



Figures 1–2: Adults of *M. gaudiella* sp. n. 1: ♂. 2: ♀.

There is some variation in the ground coloration from dull bluish to purple-violet. Sometimes there is also a small spot on the costa at 3/5. The wing pattern differs between the sexes only a little in the broadness of the fasciae, as is typical for the genus.

♂ Genitalia (Fig. 3). Uncus long, slender, with a broad rounded tip; a paired association of hair-like setae ventrally beyond the uncus; accessory claspers moderately long, nearly keel-shaped; along distal margin, a row of about 10–12 moderately long distally hook-like modified thickened setae oriented in slightly ventral direction; at inner surface proximally another irregular row of five or six shorter, finer setae; valvae moderately long, stout, distal third enlarged and strongly bent dorsally, constricted at the point of inflection; on the inner surface of the valvae a group of shorter



Figures 3–4: Genitalia of *M. gaudiella* sp. n.. 3: ♂. 4: ♀.

setae postbasally; on the inner surface of the lower margin two to three irregular rows of shorter spinous setae and some longer setae on the distal fourth part.

♀ Genitalia (Fig. 4). Tergite IX missing; sternite IX reduced, constricted medially, with strongly fringed lateral margins, without diagnostic features. Terminal papillae with two sclerotized plates forming an undiagnostic band; receptaculum seminis long and slender, constricted in the first third, with transverse striation typical for the genus.

Diagnosis. *M. trifasciella* from Piedmont (Italy) shows somewhat similar but conspicuously narrower fasciae and differs in the ground colour. *M. rablensis* from Friuli (Italy), Carinthia (Austria) and western Slovenia shows a somewhat similar wing pattern but is predominantly smaller and differs in the reddish bronzy golden to purple ground coloration of the forewings. *M. completella*, endemic to Sardinia and Corsica (Thierry and Nel 2012), has usually lighter purple-violet forewings with comparably broad fasciae and differs in the shape of the outer fascia. From all these mentioned species, the new species is furthermore clearly separated by its male genitalia, e.g. by the distinct shape of uncus and accessory claspers as well as by the orientation and form of the 10–12 long setae on the accessory claspers (Kurz and Kurz 2015). In the male genitalia, the new species closely resembles *M. isobasella* from Valais (Switzerland) (Fig. 5) and the recently described *M. stuebneri* from Sierra Nevada (Spain), being distinguished from *M. isobasella* mainly

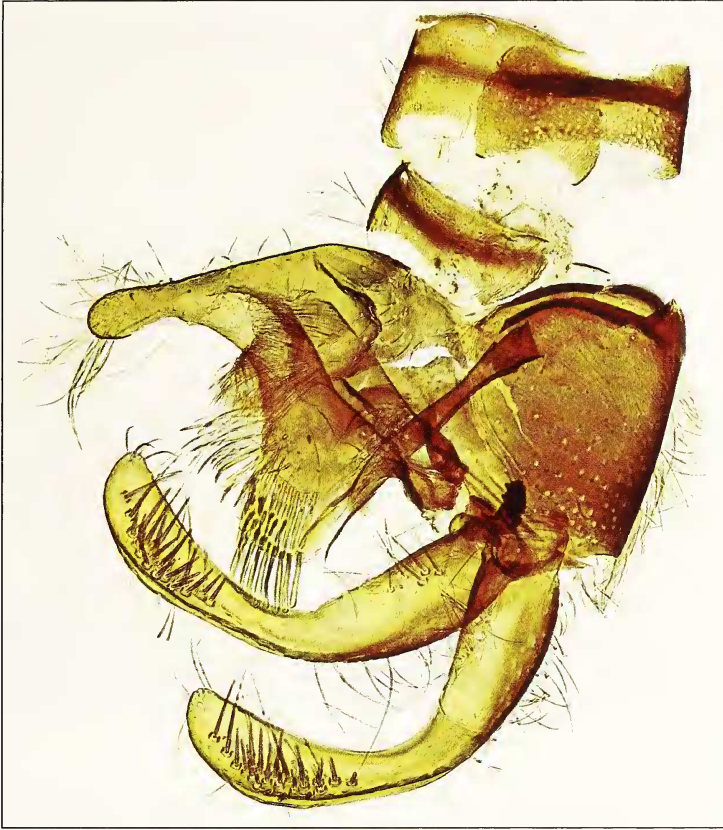


Figure 5. Male genitalia of *M. isobasella*.

by its longer uncus and the partly hook-like modified thickened setae at the rounded distal margin of the accessory claspers and from *M. stuebneri* mainly by its distinctly shorter and smaller accessory claspers (Zeller-Lukashort et al. 2013). *M. isobasella* can also easily be distinguished by its unicolorous golden forewings without any significant markings. In the female genitalia the new species differs from *M. rablensis* by the shape of sternite IX, from *M. trifasciella* and *M. comple-tella* by its strongly fringed lateral margins of sternite IX. There are no differences in the female genitalia between *M. isobasella* and the new species.

Based on morphological characters (Kurz et al. 2015), the new species is considered to belong to a species-complex together with *M. calthella*, *M. isobasella* and the recently described *M. stuebneri*.

Molecular data. The intraspecific divergence of the barcode region of *M. gaudiella* sp. n. is moderate with a mean distance of 0.46% and a maximum distance of 0.77% (n=4) (Tab. 1). The minimum distance to the nearest neighbour *M. schaefferi* on the contrary is much higher with 2.65% (mean dist. 3.18%, max. dist. 3.76%; n=9). The minimum distance to the morphologically closest *M. isobasella* is 3.67% (mean dist. 3.85%, max. dist. 4.18%; n=1) and to *M. stuebneri* 4.73% (mean dist. 4.99%, max. dist. 5.22%; n=1).

Distribution. The new species is only known from the mountain Pizzo Arero (Alpi Orobie, Piedmont, Italy) from an elevation of about 1600 m.

Table 1. Intraspecific distance and interspecific divergence to the nearest neighbour in the genus *Micropterix*, based on 25 European species. Source: DNA Barcode data from BOLD (Barcode of Life Database, cf. Ratnasingham and Hebert 2007).

Species	Mean Intra	Max intra	Nearest neighbour	Nearest species	Distance NN
<i>Micropterix allionella</i>	0.18	0.46	LEATE038-13	<i>Micropterix rothenbachii</i>	1.86
<i>Micropterix amsella</i>	N/A	N/A	LEFIG229-10	<i>Micropterix mansuetella</i>	5.35
<i>Micropterix aruncella</i>	1.7	2.99	PHLAG682-12	<i>Micropterix schaefferi</i>	2.97
<i>Micropterix aureatella</i>	0.39	0.77	PHLAG682-12	<i>Micropterix schaefferi</i>	2.74
<i>Micropterix aureoviridella</i>	0.21	0.46	PHLAG682-12	<i>Micropterix schaefferi</i>	2.74
<i>Micropterix calthella</i>	0.18	0.46	PHLAG682-12	<i>Micropterix schaefferi</i>	2.74
<i>Micropterix erctella</i>	N/A	N/A	PHLAG682-12	<i>Micropterix schaefferi</i>	2.05
<i>Micropterix facetella</i>	1.84	1.84	PHLAG682-12	<i>Micropterix schaefferi</i>	2.51
<i>Micropterix gaudiella</i>	0.46	0.77	PHLAD506-11	<i>Micropterix schaefferi</i>	2.65
<i>Micropterix hartigi</i>	N/A	N/A	PHLAG682-12	<i>Micropterix schaefferi</i>	2.74
<i>Micropterix igaloensis</i>	N/A	N/A	LEATE039-13	<i>Micropterix schaefferi</i>	2.9
<i>Micropterix isobasella</i>	N/A	N/A	PHLAG682-12	<i>Micropterix schaefferi</i>	3.01
<i>Micropterix jeanneli</i>	N/A	N/A	PHLAG682-12	<i>Micropterix schaefferi</i>	4.15
<i>Micropterix klimeschi</i>	N/A	N/A	PHLAE527-11	<i>Micropterix aruncella</i>	3.79
<i>Micropterix mansuetella</i>	N/A	N/A	LTOLB478-09	<i>Micropterix calthella</i>	4.35
<i>Micropterix myrtetella</i>	N/A	N/A	PHLAG682-12	<i>Micropterix schaefferi</i>	1.82
<i>Micropterix osthelderi</i>	0	0	PHLAG682-12	<i>Micropterix schaefferi</i>	3.21
<i>Micropterix rablensis</i>	N/A	N/A	PHLAG682-12	<i>Micropterix schaefferi</i>	2.62
<i>Micropterix rothenbachii</i>	0.21	0.32	PHLAG787-12	<i>Micropterix allionella</i>	1.86
<i>Micropterix schaefferi</i>	0.41	1.12	MICOW166-09	<i>Micropterix vulturensis</i>	0.9
<i>Micropterix sicanella</i>	N/A	N/A	PHLAG682-12	<i>Micropterix schaefferi</i>	3.44
<i>Micropterix stuebneri</i>	N/A	N/A	PHLAG682-12	<i>Micropterix schaefferi</i>	2.28
<i>Micropterix tumbergella</i>	0.05	0.16	PHLAG682-12	<i>Micropterix schaefferi</i>	3.45
<i>Micropterix vulturensis</i>	N/A	N/A	PHLAG682-12	<i>Micropterix schaefferi</i>	0.9
<i>Micropterix woceki</i>	N/A	N/A	LEATE039-13	<i>Micropterix schaefferi</i>	3.89

Life history. The early stages are unknown. The new species was observed from the end of June to mid-July near the border of a montane-subalpine beech forest with tall herbaceous vegetation and bushes (Figs 6, 7), congregating on *Rosa* sp. feeding on its pollen. Together with the new species, *M. aruncella* (Scopoli, 1763) was recognized on flowers of *Rosa* sp. too (Fig. 8). At the same locality *M. rothenbachii* Frey, 1856 was also found. Several specimens of the new species were swept from flowers of *Helianthemum* spp. in adjacent subalpine grassland. The habitat is southern exposed slopes on limestone.

Etymology. It was a great pleasure to find this unexpected new species from the Italian Alps. Therefore the new species is called “*gaudiella*”, derived from the Latin word “*gaudium*”, which means “fun, pleasure, happiness”.

Remark. The labels used by PH, Melania Massaro, Toni Mayr, Norbert Pöhl and Siegfried Ortner differ in the usage of local names of the type locality but all refer to the same spot centred on 9°47,8'E; 45°55,07'N (DDM). According to Melania Massaro there are additional specimens collected at the type locality and deposited in MBCG, which are not examined and therefore not included in the type series.



Figure 6. Biotope of *M. gaudiella* and the lucky collectors Marlies Mayr, Toni Mayr and Norbert Pöll.

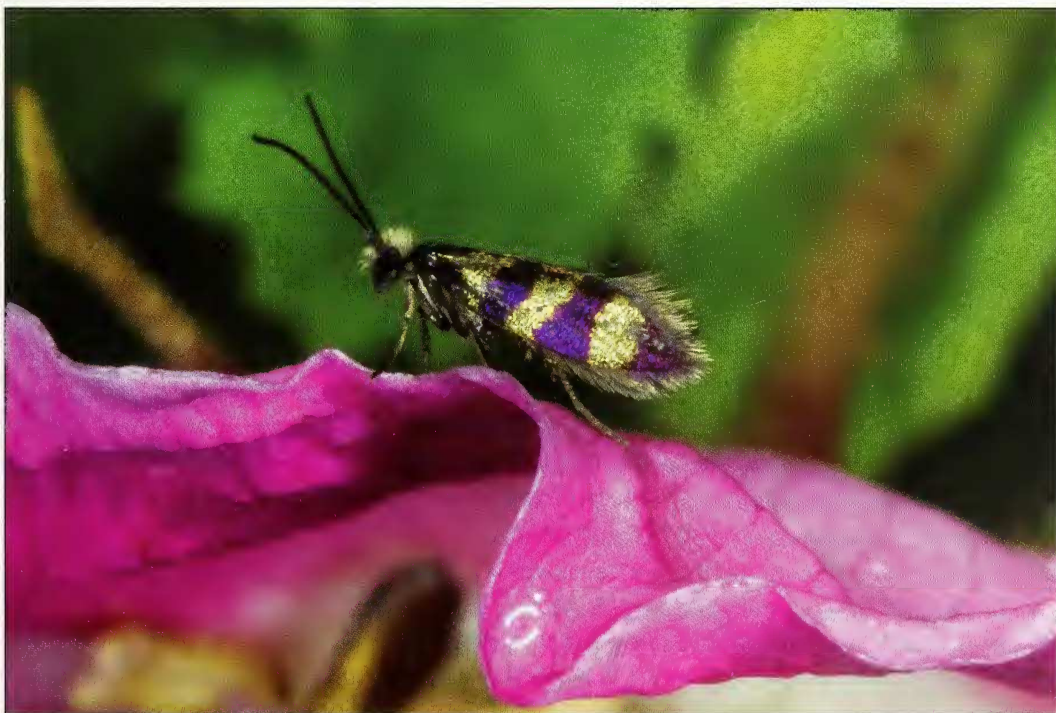


Figure 7. *M. gaudiella* sp. n., resting on a flower of *Rosa* sp..



Figure 8. *M. aruncella* and *M. gaudiella* sp. n. feeding on *Rosa* sp.

Molecular analysis

Sequencing of European species of *Micropterix* resulted in barcode fragments for 61 specimens, plus 30 unpublished or public records from external projects (e.g. Barcoding Fauna Bavarica and Finnish Barcode of Life), covering altogether 25 species, supplementing data of extra-European species that have been published by Lees et al. (2010). 74 full barcode sequences of 658 bp, 13 sequences longer than 600 bp and four sequences of ca. 400 bp were included in the analysis. The smaller fragments were single sequence only (*M. amsella*, *M. mansuetella*, and *M. rablensis*).

Barcode variation is insufficiently known for a considerable portion of species due to lack of successfully sequenced samples (Table 1). Intraspecific distance is low and ranges from 0% to 2.99% (mean 0.92%) but may include cases of overlooked cryptic diversity in species with high divergence (i.e. *M. aruncella*, *M. facetella*). Interspecific divergence for the whole sample is moderately high with mean divergence of 4.77% and maximum of 8.91%, and a mean and maximum distance to the nearest neighbour of 2.84% and 5.35% respectively. The divergence is <1% only in one species pair (*M. schaefferi* and *M. vulturensis*) (Table 1, Fig. 9).

Discussion

Alpha-taxonomy of European *Micropterix* seems quite well established with relatively few taxa described during the last decades, so it is striking to find a new species in Central Europe. By contrast, the distribution of the majority of *Micropterix* species is only moderately well documented with few

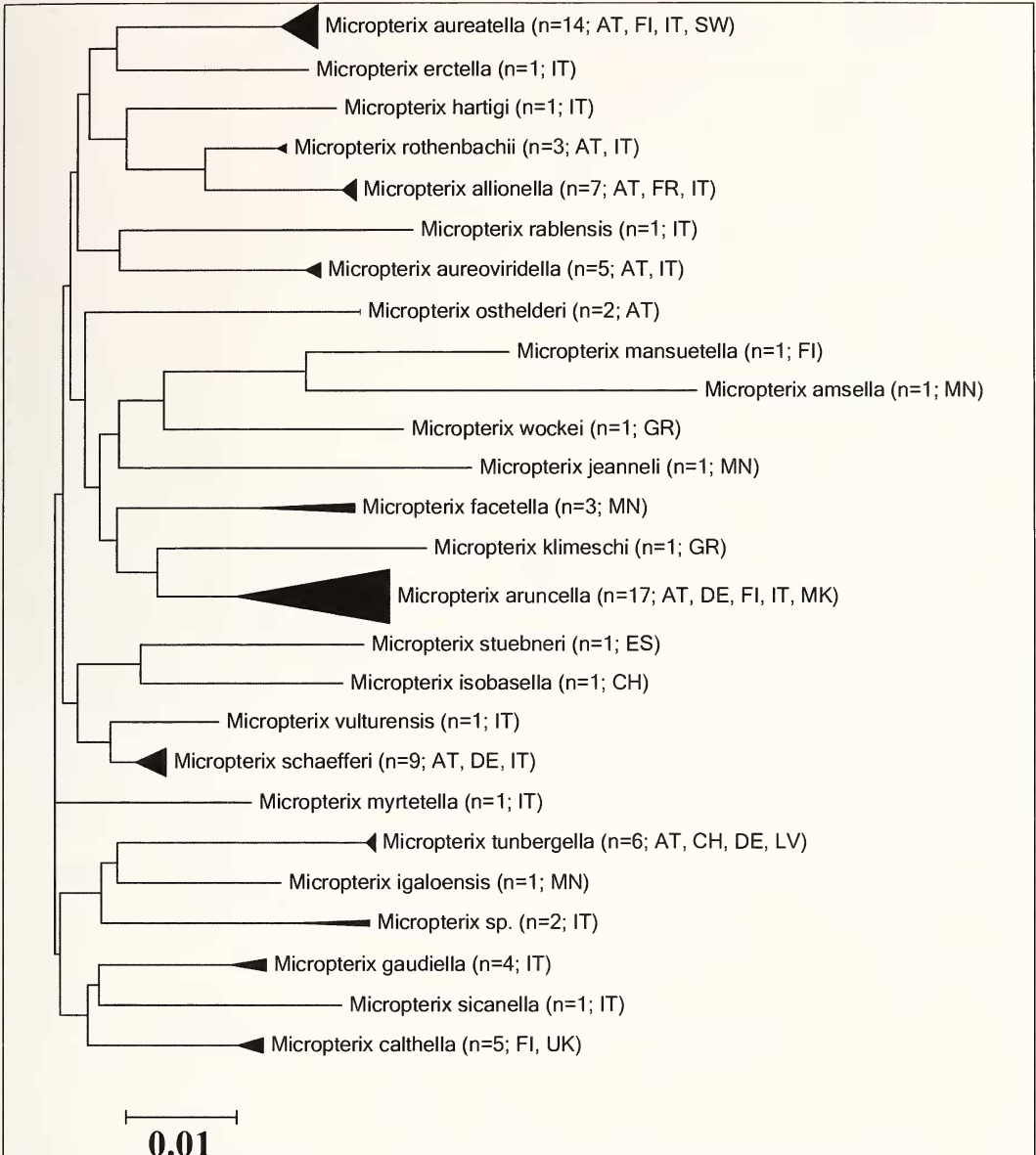


Figure 9. Neighbour-joining tree (Kimura 2-parameter, built with MEGA 5; cf. Tamura et al. 2011). Note: the scale bar only applies to internal branches between species. The width of the triangles represents the sample size, and the depth the relative genetic variation within the cluster ($2\times$ scale bar). Source: DNA Barcode data from BOLD (Barcode of Life Database, cf. Ratnasingham and Hebert 2007).

faunistic studies, particularly as concerns the Mediterranean region. Despite this shortcoming, it already appears evident from published sources that *Micropterix* exhibits remarkable levels of narrow range endemism. Indeed about three-quarters of the European taxa can be clustered into the following disjunct endemism groups (Karsholt 2015; Zeller-Lukashort et al. 2013; Kurz and Kurz 2015).

Apennines: 9 out of 20 species are endemic.

Alps: 7 out of 19 species are endemic.

Balkan Peninsula: 9 out of 17 species are endemic.

Iberian Peninsula: 5 out of 8 species are endemic.

Mediterranean islands: 8 species are endemic.

Black Sea area: 1 species is endemic.

Also, only 12 species (about one-quarter of European *Micropterix*) are widely distributed across Europe, and only one species (*M. aureatella*) reaches Japan (Kurz and Kurz 2015). *M. aruncella*, *M. aureatella*, *M. calthella*, *M. mansuetella* and *M. tunbergella* are known to range across from the mainland to geographically well separated islands (although a few micropterigids occur within an island group, such as *Sabatinca* occurring in both North and South Island of New Zealand: Gibbs 1983; Gibbs and Lees 2014; see also Imada et al. 2011). Endemism in the Lepidoptera fauna of the Alps in general and *Micropterix* in particular probably largely results from the history of glaciation, which is reflected in nine biogeographic zones defined by vegetation (Ozenda 1988). These zones have been partially considered as areas of endemism for Lepidoptera (Huemer, 1988). The majority of endemic *Micropterix* occur in the Western Alps and are restricted to the so-called “Inner Alps” (i.e. *M. isobasella*, *M. paykullella*, *M. trifasciella* and *M. fenestrellensis*) whereas *M. huemeri* is restricted to the adjacent “Pre-Ligurian” biogeographic zone. However, the highest diversity of endemic Lepidoptera in general is present in the so-called “Gardesan-Illyrian zone” of the south-eastern Alps which is defined by limestone massifs at the southern border of the Alps reaching from Lake Como in the west to the Karawanks in the east. More than 30 endemic Lepidoptera species are known from this area (Huemer 1998). *M. gaudiella* is the first species of the genus *Micropterix* probably endemic to this zone, while a further species, *M. rablensis*, is more widespread and also occurs in the northern part of the Eastern Alps. The Orobian Alps in particular seem to have attracted insufficient attention among lepidopterists so far and still host a considerable number of cryptic species belonging to various groups. A new species of *Kessleria* (Huemer & Mutanen, 2015) and a distinct subspecies of *Colostygia* (Huemer & Mayr, 2015) are striking examples. It seems not unlikely that further undescribed endemic *Micropterix* may be found in other remote and under-collected areas of Europe, e.g. on the Balkan and Iberian Peninsulas. Outside Europe, despite the efforts of John Heath, there are probably a number of undetected species in North Africa, while the Himalayas as far as China are virtually unexplored for the genus (but see Lees et al. 2010).

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